PATENT COOPERATION TREATY

From the
INTERNATIONAL SEARCHING AUTHORITY

To:

see form PCT/ISA/220

PCT

WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

(PCT Rule 43*bis*.1)

Date of mailing
(day/month/year) see form PCT/ISA/210 (second sheet)

Applicant's or agent's file reference
see form PCT/ISA/220

FOR FURTHER ACTION
See paragraph 2 below

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/EP2004/000264	15.01.2004	22.01.2003

International Patent Classification (IPC) or both national classification and IPC
C12Q1/26

Applicant
BACHER, Adelbert

1. This opinion contains indications relating to the following items:

- ☒ Box No. I Basis of the opinion
- ☒ Box No. II Priority
- ☒ Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- ☐ Box No. IV Lack of unity of invention
- ☒ Box No. V Reasoned statement under Rule 43*bis*.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- ☐ Box No. VI Certain documents cited
- ☒ Box No. VII Certain defects in the international application
- ☒ Box No. VIII Certain observations on the international application

2. **FURTHER ACTION**

If a demand for international preliminary examination is made, this opinion will usually be considered to be a written opinion of the International Preliminary Examining Authority ("IPEA"). However, this does not apply where the applicant chooses an Authority other than this one to be the IPEA and the chosen IPEA has notifed the International Bureau under Rule 66.1*bis*(b) that written opinions of this International Searching Authority will not be so considered.

If this opinion is, as provided above, considered to be a written opinion of the IPEA, the applicant is invited to submit to the IPEA a written reply together, where appropriate, with amendments, before the expiration of three months from the date of mailing of Form PCT/ISA/220 or before the expiration of 22 months from the priority date, whichever expires later.

For further options, see Form PCT/ISA/220.

3. For further details, see notes to Form PCT/ISA/220.

Name and mailing address of the ISA:

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WRITTEN OPINION OF THE INTERNATIONAL SEARCHING AUTHORITY

International application No.
PCT/EP2004/000264

Box No. I Basis of the opinion

1. With regard to the **language**, this opinion has been established on the basis of the international application in the language in which it was field, unless otherwise indicated under this item.

   ☐ This opinion has been established on the basis of a translation from the original language into the following language , which is the language of a translation furnished for the purposes of international search (under Rules 12.3 and 23.1(b)).

2. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application and necessary to the claimed invention, this opinion has been established on the basis of:

   a. type of material:

   ☒ a sequence listing

   ☐ table(s) related to the sequence listing

   b. format of material:

   ☒ in written format

   ☒ in computer readable form

   c. time of filing/furnishing:

   ☐ contained in the international application as filed.

   ☐ filed together with the international application in computer readable form.

   ☒ furnished subsequently to this Authority for the purposes of search.

3. ☒ In addition, in the case that more than one version or copy of a sequence listing and/or table relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

4. Additional comments:

Box No. II Priority

1. ☒ The following document has not been furnished:

   ☒ copy of the earlier application whose priority has been claimed (Rule 43*bis*.1 and 66.7(a)).

   ☐ translation of the earlier application whose priority has been claimed (Rule 43*bis*.1 and 66.7(b)).

   Consequently it has not been possible to consider the validity of the priority claim. This opinion has nevertheless been established on the assumption that the relevant date is the claimed priority date.

2. ☐ This opinion has been established as if no priority had been claimed due to the fact that the priority claim has been found invalid (Rules 43*bis*.1 and 64.1). Thus for the purposes of this opinion, the international filing date indicated above is considered to be the relevant date.

3. Additional observations, if necessary:

Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non obvious), or to be industrially applicable have not been examined in respect of:

☐ the entire international application,

☒ claims Nos. 11

because:

☐ the said international application, or the said claims Nos. relate to the following subject matter which does not require an international preliminary examination *(specify)*:

☐ the description, claims or drawings *(indicate particular elements below)* or said claims Nos. are so unclear that no meaningful opinion could be formed *(specify)*:

☐ the claims, or said claims Nos. are so inadequately supported by the description that no meaningful opinion could be formed.

☒ no international search report has been established for the whole application or for said claims Nos. 11

☐ the nucleotide and/or amino acid sequence listing does not comply with the standard provided for in Annex C of the Administrative Instructions in that:

the written form	☐	has not been furnished
	☐	does not comply with the standard
the computer readable form	☐	has not been furnished
	☐	does not comply with the standard

☐ the tables related to the nucleotide and/or amino acid sequence listing, if in computer readable form only, do not comply with the technical requirements provided for in Annex C-*bis* of the Administrative Instructions.

☐ See separate sheet for further details

Box No. V Reasoned statement under Rule 43*bis*.1(a)(i) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1: Statement

Novelty (N)	Yes:	Claims	2-8
	No:	Claims	1,9,10
Inventive step (IS)	Yes:	Claims	2-6
	No:	Claims	1,7-10
Industrial applicability (IA)	Yes:	Claims	1-10
	No:	Claims	

2. Citations and explanations

see separate sheet

Box No. VII Certain defects in the international application

The following defects in the form or contents of the international application have been noted:

see separate sheet

Box No. VIII Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

see separate sheet

Reference is made to the following documents:

D1: WO 02/083720 A (EISENREICH WOLFGANG ; ROHDICH FELIX (DE); ADAM PETRA (DE); BACHER ADEL) 24 October 2002 (2002-10-24)

D2: ALTINCICEK B ET AL: "LytB protein catalyzes the terminal step of the 2-C-methyl-D-erythritol-4-phosphate pathway of isoprenoid biosynthesis" FEBS LETTERS, ELSEVIER SCIENCE PUBLISHERS, AMSTERDAM, NL, vol. 532, no. 3, 18 December 2002 (2002-12-18), pages 437-440, XP004398446 ISSN: 0014-5793

D3: ADAM PETRA ET AL: "Biosynthesis of terpenes: studies on 1-hydroxy-2-methyl-2-(E)-butenyl 4-diphosphate reductase." PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA. 17 SEP 2002, vol. 99, no. 19, 17 September 2002 (2002-09-17), pages 12108-12113, XP002289308 ISSN: 0027-8424

D4: OLLAGNIER S ET AL: "Activation of the anaerobic ribonucleotide reductase from Escherichia coli: The essential role of the iron-sulfur center for S-adenosylmethionine reduction" JOURNAL OF BIOLOGICAL CHEMISTRY, vol. 272, no. 39, 1997, pages 24216-24223, XP002289404 ISSN: 0021-9258

Re Item III

Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. Present claim 11 relates to a compound defined by reference to a desirable characteristic or property, namely its identification of being an inhibitor via one of several claimed assays. The claim covers all compounds having this characteristic or property, whereas the application provides no support within the meaning of Article 6 PCT and no disclosure within the meaning of Article 5 PCT for such compounds.
   Independent of the above reasoning, the claim also lacks clarity (Article 6 PCT), because an attempt is made to define the compound by reference to a result to be achieved.
   Hence, no meaningful opinion can be formed on the novelty, inventive step and industrial applicability of the claimed invention (Article 34(4)(a)(ii) PCT).

Re Item V

Reasoned statement with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. The present application does not meet the criteria of Article 33(1) PCT, because the subject-matter of independent claim 1 is not new in the sense of Article 33(2) PCT.

   The document D1 discloses (the references in parentheses applying to this document)
   an assay for testing a sample for the presence or absence of inhibition of the enzymatic conversion of 1-hydroxy-2-methyl-(E)-butenyl 4-diphosphate into isopentyl diphosphate and dimethylallyl diphosphate (page 20, line 15-18) by the steps
   a) reacting an aqueous mixture containing 1-hydroxy-2-methyl-(E)-butenyl 4-diphosphate (page 20, line 20), a 1-hydroxy-2-methyl-(E)-butenyl 4-diphosphate reductase (page 20, line 19), and a reducing agent (page 20, line 27-28: NADH or NADPH; line 22: FAD) under predetermined reaction conditions for a predetermined period of time (page 20, line 19-22);
   b) analyzing the reaction mixture obtained in step a) for the consumed amount of 1-hydroxy-2-methyl-(E)-butenyl 4-diphosphate (page 20, line 23) and/or NAD(P)H (page 20, line 27-28) and/or for the produced amount of isopentyl diphosphate (page 20, line 24) and/or dimethylallyl diphosphate (page 20, line 24);
   c) repeating step a) in the presence of the sample to be tested (page 20, line 21);
   d) repeating step b) with the reaction mixture defined in step c) (page 20, line 23-24);
   e) comparing the results of steps b) and d) (page 20, line 25-26).

   The subject-matter of independent claim 1 is therefore not new (Article 33(2) PCT).

2. Dependent claims 7-10 do not contain any features which, in combination with the features of any claim to which they refer, meet the requirements of the PCT in respect of novelty and/or inventive step, because the features are either known from documents D1-D3 or are within the scope of the normal experience and

competence of a skilled person.

3. The subject-matter of independent claim 2 is new in the sense of Article 33(2) PCT and inventive in the sense of Article 33(3) PCT for the following reasons:

Document D1 is regarded as being the closest prior art to the subject-matter of claim 2, and discloses (the references in parentheses applying to this document):

an assay according to claim 1 of the present application (see above).

The subject-matter of claim 2 therefore differs from this known from document D1 in the use of the NAD(P)H/flavodoxin/flavodoxin reductase system instead of NAD(P)H and FAD.

The subject-matter of claim 2 is therefore new (Article 33(2) PCT).

The problem to be solved by the present invention may be regarded as how to provide the 1-hydroxy-2-methyl-(E)-butenyl 4-diphosphate reductase with electrons in an alternative way.

The solution proposed in claim 2 of the present application is the use of the NAD(P)H/flavodoxin/flavodoxin reductase system.

This has to be considered as involving an inventive step (Article 33(3) PCT). Even if the person skilled in the art knows from D4 that the NAD(P)H/flavodoxin/flavodoxin reductase system can be used as an alternative to chemical reducing systems for the provision of a reductase with electrons, there is no hint that it would work for the present enzyme, too. Particularly with regard to the fact that D5 describes the generation of a glycil radical in the reductase whereas no details are known about the mechanism of action of the 1-hydroxy-2-methyl-(E)-butenyl 4-diphosphate reductase. Hence, the subject-matter of claim 2 is inventive (Article 33(3) PCT).

4. Claims 3-6 are dependent on claim 2 and as such also meet the requirements of the PCT with respect to novelty and inventive step.

5. Claims 1-10 appear to meet the requirements of the PCT with respect to the

industrial applicability of their subject-matter (Article 33(4) PCT).

Re Item VII
Certain defects in the international application

1. Contrary to the requirements of Rule 5.1(a)(I) PCT, the technical field to which the invention relates is not specified in the description.

2. Contrary to the requirements of Rule 5.1(a)(ii) PCT, the relevant background art disclosed in the document D3 is not mentioned in the description, nor is this document identified therein. In addition, the relevant background art disclosed in the document D2 is not mentioned in the description even if the document is identified therein.

3. Contrary to the requirements of Rule 5.1(a)(iv) PCT, the figures in the drawings are not briefly described in the description.

4. Claim 6 erroneously refers to itself.

Re Item VIII
Certain observations on the international application

1. The wording of claim 4 is unclear and leaves the reader in doubt as to the meaning of the technical features to which it refers, thereby rendering the definition of the subject-matter of said claim unclear, Article 6 PCT: If the produced amount of NAD(P)+ or isopentenyl diphosphate and/or dimethylallyl diphosphate is **to be tested exclusively** and the consumed amount of NAD(P)H or HMBPP is **not to be tested**, then claim 4 can't be dependent on claim 3. However, if the testing of the produced amount of the compounds just mentioned does not exclude the testing of the consumed amount of the compounds just mentioned, claim 4 is inherent in claim 2 and should be deleted.